

17. A method for determining the sequence of a nucleic acid molecule using a mutant RNA polymerase which has a reduced discrimination for non-canonical versus canonical nucleotides as substrates, comprising the steps of:

5           a) synthesizing a nucleic acid molecule *de novo* from an RNAP promoter sequence in a reaction mixture containing the mutant RNA polymerase in each of four separate reactions, each reaction comprising at least four nucleoside triphosphates, wherein at least one  
10       nucleoside triphosphate has a nucleic acid base which is complementary to each of adenine, cytidine, guanine and uracil or thymine and a sugar with either a hydroxy or a hydrogen or a fluorine at the 2'-position, and a portion of a ddNTP, such that each of the four separate  
15       reactions contains a ddNTP that is complementary to a different one of the four common nucleic acid bases in a nucleic acid molecule, and

          b) evaluating the reaction products so that the sequence of the template molecule may be deduced.

18. The method of claim 17 wherein, each reaction comprises at least four nucleoside triphosphates chosen from the group consisting of ATP, CTP, GTP, and UTP or rTTP, and further comprises one ddNTP.

19. The method of claim 17 wherein, each reaction comprises at least four nucleoside triphosphates chosen from the group consisting of dATP, dCTP, dGTP, dUTP, dTTP, 7-deaza-dGTP, dITP, 5-methyl-dCTP, 5-hydroxy-methyl-dCTP, and  
5       further comprises one ddNTP.

20. The method of claim 17 wherein, each reaction comprises at least four nucleoside triphosphates chosen from the group consisting of 2'-F-ATP, 2'-F-CTP, 2'-F-GTP, 2'-F-UTP, 2'-F-TTP, 2'-deaza-2'-F-GTP, 2'-F-ITP, and 5-methyl-2'-F-CTP, 5-hydroxymethyl-2'-F-CTP and further comprises one ddNTP.

21. A method for determining the sequence of a nucleic acid molecule using a mutant RNA polymerase which has a reduced discrimination for non-canonical versus canonical nucleotides as substrates, comprising the steps of:

- 5           a) synthesizing a nucleic acid molecule by extending a primer, wherein at least part of the primer is complementary to a template molecule so as to anneal therewith, in a reaction mixture containing the mutant RNA polymerase in each of four separate reactions, each  
10           reaction comprising at least four nucleoside triphosphates, wherein at least one nucleoside triphosphate has a nucleic acid base which is complementary to each of adenine, cytidine, guanine and uracil or thymine and a sugar with either an hydroxy or  
15           a hydrogen or a fluorine at the 2'-position, and further comprising a portion of a ddNTP, such that each of the four separate reactions contains a ddNTP that is complementary to a different one of the four common nucleic acid bases, and
- 20           b) evaluating the reaction products so that the sequence of the template molecule may be deduced.

28. The method of claim 25, wherein the dinucleotide or trinucleotide in the reaction mixture is modified to contain a radioactive or non-radioactive label.

29. A kit for performing a dideoxy sequencing reaction, comprising a mutant nucleic acid polymerase which has reduced discrimination between canonical and non-canonical nucleoside triphosphates and data or information  
5 describing conditions under which the method may be performed.

30. A method for determining the sequence of a nucleic acid molecule using a mutant RNA polymerase which has a reduced discrimination for non-canonical versus canonical nucleotides as substrates, comprising the steps of:

5 a) synthesizing a nucleic acid molecule *de novo* from an RNAP promoter sequence in a reaction mixture containing a mutant RNA polymerase in each of four separate reactions, each reaction comprising at least four nucleoside triphosphates, wherein at least one  
10 nucleoside triphosphate has a nucleic acid base which is complementary to each of adenine, cytidine, guanine and uracil or thymine and a sugar with a hydrogen or a fluorine at the 2'-position, and a portion of a rNTP, such that each of the four separate reactions contains  
15 a rNTP that is complementary to a different one of the four common nucleic acid bases, and

b) treating the nucleic acid products of the reactions so as to bring about hydrolysis of the phosphodiester backbone at all sites where a  
20 ribonucleotide has been incorporated, and